Application Serial No. 09/063,356 Page 2 August 27, 1998

second binding reagent immobilized on the walls of at least a second group of said channels, and

channels), (B) detecting binding between a binding target in the sample and at least one binding reagent on the walls of at least one group of discrete channels in the substrate, thereby detecting said binding reaction.

22. A method according to claim 21, wherein said substrate is fabricated from glass or silicon.

23. A method according to claim 22, wherein said substrate is made of nanochannel

glass.

24. A method according to claim 22, wherein said substrate is made of oriented array microporous silicon.

- 25. A method according to claim 21, wherein the first and second binding reagents differ from one another.
- 26. A method according to claim 21, wherein the first and second binding reagents bind different binding targets.
- 27. A method according to claim 21, wherein said device comprises discrete channels having diameters of from about 0.033 micrometers to about 10 micrometers.
- 28. A method according to claim 21, wherein said device comprises discrete channels having diameters of from about 0.45 micrometers to about 10 micrometers.
- 29. A method according to claim 21, wherein said device comprises discrete channels having cross sectional areas of between about  $8.5 \times 10^{-4} \ \mu m^2$  to about  $80 \ \mu m^2$ .

-2-

- 30. A method according to claim 21, wherein said substrate is between about 100  $\mu m$  to about 1000  $\mu m$  thick.
- 31. A method according to claim 21, wherein said substrate comprises channels have an inner surface area of between about 10  $\mu m^2$  and about 3 X 10<sup>4</sup>  $\mu m^2$ .
- 32. A method according to claim 21, wherein said substrate comprises groups of channels having areas of between about 20  $\mu m^2$  to about 3 X  $10^6~\mu m^2$ .
- 33. A method according to claim 21, wherein there are between 400 and 4400 of said groups of discrete channels per cm<sup>2</sup> of cross-sectional area of the substrate.
- 34. A method according to claim 21, wherein the inner surface area of the channels in a group of channels of said substrate is from about 100 to about 1000 times the cross sectional area of the group of channels.
  - 35. A method according to claim 29, 31, 32, or 34, wherein said substrate is fabricated from glass or silicon.
  - 36. A method according to claim 35, wherein said substrate is made of nanochannel glass or oriented array microporous silicon.
  - 37. A method according to claim 21 wherein a detectable label is used to detect the binding reaction.
  - 38. A method according to claim 36, wherein said detectable label is selected from the group consisting of fluorescent, chemiluminescent and radioactive labels.
  - 39. A method according to claim 38, wherein said detectable label is attached to said binding target.

-3-002.149004

- A method according to claim 21, 23, 24, 25, 29, 31, 32, 34, or 37, wherein said binding reagents are effective for carrying out binding reactions selected from the group consisting of binding reactions involving small molecules, macromolecules, particles and cellular systems.
  - A method according to claim 40, wherein said binding reagents are effective for carrying out an analytical task selected from the group consisting of sequence analysis by hybridization, immunochemical analysis of protein mixtures, epitope mapping, assay of receptor-ligand interactions and profiling of cellular populations involving binding of cell surface molecules to specific ligands or receptors.
    - A method according to claim 41, wherein said binding reagents are selected 42. from the group consisting of DNA, proteins and ligands.
    - A method according to claim 42, wherein said binding reagents are 43. oligonucleotide probes.

A method for detecting expression of at least one gene, comprising:

- contacting a sample suspected of containing a binding target indicative of 44. gene expression with a substrate comprised of: (i) oppositely facing first and second major surfaces, (ii) a multiplicity of discrete channels extending through said substrate from said first major surface to said second major surface, (iii) at least a first binding reagent for detecting expression of a gene, wherein said first binding reagent is immobilized on the walls of at least a first group of said channels, and (iv) at least a second binding reagent for detecting expression of a gene, wherein said second binding reagent is immobilized on the walls of at least a second group of channels, and,
  - detecting binding between at least one binding target indicative of gene expression in the sample and at least one binding reagent on the walls of at least one group of discrete channels in the substrate, thereby detecting the expression of at least one gene.

-4-

8

- 45. A method according to claim 44, wherein said substrate is fabricated from glass or silicon.
- 46. A method according to claim 45, wherein said substrate is made of nanochannel glass.
  - 47. A method according to claim 45, wherein said substrate is made of oriented array microporous silicon.
  - 48. A method according to claim 44, wherein the first and second binding reagents differ from one another.
  - 49. A method according to claim 44, wherein the first and second binding reagents bind different binding targets.
  - 50. A method according to claim 44, wherein said device comprises discrete channels having cross sectional areas of between about  $8.5 \times 10^{-4} \ \mu m^2$  to about  $80 \ \mu m^2$ .
  - 51. A method according to claim 44, wherein said substrate comprises channels have an inner surface area of between about 10  $\mu m^2$  and about 3 X  $10^4$   $\mu m^2$ .
  - 52. A method according to claim 44, wherein said substrate comprises groups of channels having areas of between about 20  $\mu m^2$  to about 3 X  $10^6~\mu m^2$ .
  - 53. A method according to claim 44, wherein there are between 400 and 4400 of said groups of discrete channels per cm<sup>2</sup> of cross-sectional area of the substrate.
  - 54. A method according to claim 44, wherein the inner surface area of the channels in a group of channels of said substrate is from about 100 to about 1000 times the cross sectional area of the group of channels.

-5-

002.149004

August 27, 1998

- A method according to claim 44, wherein a detectable label is used to detect the 55. binding reaction.
- A method according to 55, wherein said detectable label is selected from the group consisting of fluorescent, chemiluminescent and radioactive labels. 56.
- A method according to claim 56, wherein said detectable label is attached to said 57. binding target.
- A method according to claim 44, wherein said binding reagents bind to said 58. binding targets by hybridization.
- A method according to 58, wherein said binding targets indicative of gene 59. expression are RNA or cDNA molecules.
- A method according to claim 59, wherein said binding reagents are 60. polynucleotides.
- A method according to claim 52, wherein the wall of said groups of channels each have immobilized thereon at least one binding reagent that can hybridize to a binding target, wherein said binding target is a molecule indicative of expression of at least one gene.
- A method according to claim 61, wherein the binding reagent in each group of channels comprises a probe for detecting differences in gene expression between samples.
- A method according to claim, 62, wherein said probes detect differences in 63. gene expression in cells subjected to different conditions.
  - A method according to claim 63, wherein said probes detect differences in 64. gene expression between normal and mutated states of a cell or tissue.

Application Serial No. 09/063,356 Page 7 August 27, 1998

65. A method according to claim 64, wherein said probe detects differences in gene expression of cells caused by exposure to a drug or chemical compound.

A method for detecting a sequence variation in at least one gene, comprising:

(A) contacting a sample suspected of containing a binding target indicative of a sequence variation in at least one gene with a substrate comprised of: (i) oppositely facing first and second major surfaces, (ii) a multiplicity of discrete channels extending through said substrate from said first major surface to said second major surface, and (iii) at least a first binding reagent for detecting a variation of a gene immobilized on the walls of at least a first group of said channels and (iv) at least a second binding reagent for detecting a variation of a gene immobilized on the walls of at least a second group of channels,

- (B) detecting binding between at least one binding targets indicative of sequence variation in the sample and at least one binding reagent on the walls of at least one group of discrete channels in the substrate, thereby detecting the sequence variation in at least one gene.
- 67. A method according to claim 66, wherein said substrate is fabricated from glass or silicon.
- 68. A method according to claim 67, wherein said substrate is made of nanochannel glass.
- 69. A method according to claim 67, wherein said substrate is made of oriented array microporous silicon.
- 70. A method according to claim 66, wherein the first and second binding reagents differ from one another.
- 71. A method according to claim 66, wherein the first and second binding reagents bind different binding targets.

CONT

- 72. A method according to claim 66, wherein said device comprises discrete channels having cross sectional areas of between about  $8.5 \times 10^{-4} \, \mu m^2$  to about  $80 \, \mu m^2$ .
- 73. A method according to claim 66, wherein said substrate comprises channels have an inner surface area of between about 10  $\mu m^2$  and about 3 X  $10^4$   $\mu m^2$ .
- 74. A method according to claim 66, wherein said substrate comprises groups of channels having areas of between about 20  $\mu m^2$  to about 3 X 10<sup>6</sup>  $\mu m^2$ .
- 75. A method according to claim 66, wherein there are between 400 and 4400 of said groups of discrete channels per cm<sup>2</sup> of cross-sectional area of the substrate.
- 76. A method according to claim 66, wherein the inner surface area of the channels in a group of channels of said substrate is from about 100 to about 1000 times the cross sectional area of the group of channels.
- 77. A method according to claim 66, wherein a detectable label is used to detect the binding reaction.
- 78. A method according to claim 77, wherein said detectable label is selected from the group consisting of fluorescent, chemiluminescent and radioactive labels.
- 79. A method according to claim 78, wherein the label is attached to the binding target.
- 80. A method according to claims 66 or 74, wherein said binding reagents bind to said binding targets by hybridization.
- 81. A method according to 80, wherein said target molecules are selected from the group consisting of RNA, cDNA and genomic DNA molecules.

Marca